

**EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION
ORGANISATION EUROPEENNE ET MEDITERRANEENNE POUR LA PROTECTION
DES PLANTES**

(11-17239)

Summary sheet of validation data for a diagnostic test

The EPPO Standard PM 7/98 *Specific requirements for laboratories preparing accreditation for a plant pest diagnostic activity* describes how validation should be conducted. It also includes definitions of performance criteria.

Target Organism	Bursaphelenchus xylophilus	
Short description	Detection of Bursaphelenchus xylophilus in wood extract with real-time PCR Leal et al. 2005	
Laboratory contact details	Anses, Laboratoire de la Santé des Végétaux - Unité de Nématologie Domaine de la Motte au Viconte BP 35327, 35653 Le Rheu, France	
Date and reference of the validation report	2011-02 - Anses 2011 Rapport d'évaluation d'outils moléculaires de détection de Bursaphelenchus xylophilus sur extrait de bois	
Validation process according to EPPO Standard PM 7/98:	Yes	
Reference of the test description	0 Leal I, Green M, Allen E, Humble L, Rott M (2005) An effective PCR-based diagnostic method for the detection of Bursaphelenchus xylophilus in wood samples from lodgepole pine. Nematology Vol.7(6), 833-842	
Is the test the same as described in the EPPO DP?	No Other test	
Is the lab accredited for this test?	No	
Plant species tested (if relevant)		
Matrices tested (if relevant)	Wood extract	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix		
Molecular methods, e.g. hybridization, PCR and real time PCR	X	real time PCR (Taqman)

Serological methods: IF, ELISA, Direct Tissue Blot Immuno Assay		
Plating methods: selective isolation		
Bioassay methods: selective enrichment in host plants, baiting, plant test and grafting.		
Pathogenicity test		
Fingerprint methods: protein profiling, fatty acid profiling & DNA profiling		
Morphological and morphometrical methods intended for identification		
Biochemical methods: e.g. enzyme electrophoresis, protein profiling		
Other		
<u>Analytical sensitivity (= limit of detection)</u>		
What is smallest amount of target that can be detected reliably?	one nematode	
<u>Diagnostic sensitivity</u>		
Proportion of infected/infested samples tested positive compared to results from the standard test , see appendix 2 of PM 7/98	100%	
Specify the standard test	morphology	
<u>Analytical specificity</u>		
Specificity value	100%	
Number of strains/populations of target organisms tested	7 populations (see table1)	
Number of non-target organisms tested	15 populations (see table1)	
Cross reacts with (specify the species)	none	
<u>Diagnostic Specificity</u>		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	99% (6 false positives results/490 uninfested samples and 1 false negative result /13 infested samples)	
Specify the standard test	morphology	
<u>Reproducibility</u>		

Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% for one single nematode
<u>Repeatability</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% for one single nematode
<u>Test performance study</u>	
Test performance study?	No
Include brief details of the test performance study and its output. If available, provide a link to published article/report	
<u>Other information</u>	
Any other information considered useful e.g. robustness, ease of performing the test, etc.	The full report is available upon request to the laboratory
The following complementary files are available online:	<ul style="list-style-type: none"> • Table 1 List of species and population tested • Table 2 comparison of different PCR tests B xylophilus identification